REMARKS

IN THE CLAIMS

Claims 1, 2, 5, 9 and 10 have been rejected under 35 U.S.C. § 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which the Applicants regard as the invention.

Claim 1 has been amended by introducing the word "the" between "consisting of" and "sequences set forth".

Claim 2 has been deleted from the application.

Claim 5 has been amended by limiting the group of plant to alfalfa.

Claim 10 has been amended by limiting the transgenic expression of a foreign DNA of interest to leaves of the plant cells or plants.

According to the amendments introduced into claims 1, 5 and 10, Applicants believe that the expression of a foreign DNA of interest into plants or plant cells, now being limited to leaves, is in accordance with the state of the art, particularly with Oommenn et al. (1994, The Plant Cell, vol. 6: 1789-1803) cited in the Office Action dated June 3, 2003.

No new matter has been added with the amendment.

IN THE SPECIFICATION

In the first and second lines of the specification, the sentence "this application is a continuation application of US serial number 09/678,303 filed on October 3, 2000" has been deleted. As a petition to revive the present application number 09/678,303 has been granted on June 6, 2002, paper number 8, it is respectfully submitted that the present application be prosecuted as such, and not as a continuation of a parent application.

The Examiner has rejected the specification under 35 U.S.C. § 112, first paragraph, as containing subject matter which was not described in the specification in such a way as to enable one skilled in the art to which it pertains, or with which it is most nearly connected to make and/or use the invention.

A plurality of amendments were carried out in the specification by the Applicants to clarify and formalize the application.

In the paragraph bridging on line 30 of page 4 to line 9 of page 5, the disclosure was amended to replace the claim language, namely to replace the term "various" by "SEQ ID NO:2 and SEQ ID NO:3" which are described in the application and are two deletion fragments of "SEQ ID NO:1", the term SEQ ID NO:1 replaces the occurrence of "pGPlas3.2".

By limiting the deletion fragments to SEQ ID NO:2 and 3, which are fragments of SEQ ID NO:1, which themselves replaces the term pGPlas3.2, Applicants believe that the specification is now in condition enabling someone skilled in the art to carry out the invention as claimed in claim 1, 5, 9 and 10. Claim 2 was deleted from the application. No new matter has been added.

Applicants respectfully submit that the specification and claims as amended render the application in condition for allowance. Reconsideration of the objections is respectfully requested. In the event that there are any questions concerning this amendment or the application in general, the Examiner is respectfully urged to telephone the undersigned so that persecution of this application may be expedited.

Respectfully submitted,

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CERTIFICATE OF FACSIMILE TRANSMISSION

I hereby certify that this paper is being facsimile transmitted to the Patent and Trademark Office on the date shown below.

Paul Marcoux , 24,990
Name of person signing certification

Signature

September 24, 2003
Date

PROMOTER FOR REGULATING EXPRESSION OF FOREIGN GENES

~~This applicatioin is a continuation application of US Serial No. 09/678,303 filed on October 3, 2000.~~

BACKGROUND OF THE INVENTION

(a) Field of the Invention

The invention relates to a promoter for regulating expression of foreign genes in a transgenic organism, more specifically in a leaf-specific manner in transgenic plants.

(b) Description of Prior Art

Genetic transformation of microbes have been used for more than 15 years to produce useful recombinant molecules, and applications in the pharmaceutical, cosmaceutical and dermaceutical industries are being currently exploited. This technology has expanded from microbes to plants and animals in the last ten years with the development of techniques required to adapt this general concept to complex eukaryotic organisms. Basically a gene encoding for a protein of interest or a gene encoding for an enzyme responsible for a modification of a metabolic pathway that leads to a molecule of interest, is linked in an appropriate fashion to cis-and trans-acting regulatory sequences, and transferred to a target cell where it is incorporated in the molecular machinery (in a transitory or stable fashion). The transgenic cell, or a tissue or organism regenerated from the transgenic cell will then perform transcription and translation of the transgene and therefore be enabled to accumulate the protein of interest or to perform the new metabolic reaction through the activity of the enzyme of interest.

The emerging industry of molecular farming is one of the most promising industry of the coming century. Its promise is to provide safe and renewable molecule factories for the industry. Among the applications that are currently developed are the production of low-cost monoclonal antibodies for therapeutic and diagnostic uses, the production of unlimited amounts of hormones, cytokines and other bio-active molecules for the treatment of chronicle or lethal diseases, the production of bio-safe substitutes for various blood components, the production of unlimited amounts of processing enzymes for the food and pulp industry, the production of low-cost enzymes for waste treatments, and the production of safe bio-active molecules for the cosmetic industry.

ORF of a gene, wherein said promoter is operationally located with respect to said gene for expression of said gene.

For the purpose of the present invention the following terms are defined below.

The expression "functional fragments or derivatives thereof" is intended to mean any derivative or fragment of sequences SEQ ID NOS:1-3 which allow for an equivalent level of expression of a foreign gene as the promoter of the present invention set forth in SEQ ID NOS:1-3.

DETAILED DESCRIPTION OF THE INVENTION

Following is a detailed description of the method used to generate transgenic alfalfa lines that can be regulated in their expression of a reporter gene.

In this embodiment, a promoter having the sequence set forth in SEQ ID NOS:1-3 was then ligated to a reporter gene and a terminator, and this construct was inserted in suitable plant expression vectors for DNA bombardment onto alfalfa leaves and for *Agrobacterium* mediated DNA transfer as described by Desgagnés et al. (1995, *Plant Cell Tissue Organ Cult.* 42:129-140). These two DNA transfer methods were used to demonstrate that expression of the reporter gene can be modulated by light.

Materials and Methods

DNA sequencing

DNA sequencing was performed as described by Sanger et al (1977, *P.N.A.S. USA*, 74:5643-5647).

The resulting promoters of the present invention have the sequence as set forth in SEQ ID NOS: 1 to 3.

Construction of expression cassettes and vectors

The cassettes for expression analysis using the GUS reporter gene were assembled as follows. A promoterless GUS cassette was digested from pBI101 with HindIII and EcoRI, and was inserted into the HindIII and EcoRI sites of the pUC19 polycloning site. The resulting plasmid was named pBI201

and was used for further constructs. ~~Various~~ SEQ ID NO:2 and SEQ ID NO:3, two and deletion fragments of ~~pGPlas3.2~~ SEQ ID NO:1, were operably ~~transcriptionally and transitionally~~ fused at the 5'terminus of the GUS reporter gene in pBI201 by PCR ligation, and ~~these~~ resulting constructs were used for transitory expression studies using DNA bombardment. ~~Upon identification of the adequate deletion fragment, it was~~ or subcloned into a binary plant expression vector such as pBI101 (Clonetech). These recombinant plasmids were used for stable integration through *A. tumefaciens* infection as described below.

Agrobacterium-mediated DNA transfer and regeneration of transgenic lines

The recombinant plasmids were introduced into *Agrobacterium tumefaciens* strain LBA4404 by electroporation as described in Khoudi et al (1999, *Biotechnol. Bioeng.*, 64:135-143). Selected *Agrobacterium* strains were then co-cultivated with leaf disks from genotype C5-1 for 4 days in the absence of selection pressure (kanamycin). Following this incubation period, leaf disks were washed and pampered, and then allowed to form calli onto medium B5H. Calli were then transferred for 21 days on SH medium for embryo induction and for 28 days on BOi2Y for embryo development. Torpedo-shaped embryos were removed from Boi2Y and placed on MS medium for regeneration. Kanamycin was present in all cultivation medium except for co-cultivation and regeneration on MS. This method is described in length in Desgagnés et al (1995, *Plant Cell Tissue Organ Cult.* 42:129-140). Rooted plantlets were grown to maturity in the greenhouse.

While the invention has been described in connection with specific embodiments thereof, it will be understood that it is capable of further modifications and this application is intended to cover any variations, uses, or adaptations of the invention following, in general, the principles of the invention and including such departures from the present disclosure as come within known or customary practice within the art to which the invention pertains and as may be applied to the essential features hereinbefore set forth, and as follows in the scope of the appended claims.

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